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Increased serum adenosine deaminase activity in schizophrenic receiving antipsychotic treatment

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Abstract

Adenosine is an important modulator of the nervous system that has been implicated in the pathophysiology of schizophrenia. We studied peripheral adenosine metabolism by determining the activity of serum adenosine deaminase, which converts adenosine into inosine, and 5′-nucleotidase, which converts AMP into adenosine, in 26 DSM-IV male schizophrenic patients under antipsychotic monotherapy and 26 healthy volunteers balanced for age and race. Schizophrenic patients treated either with typical antipsychotics or clozapine showed increased serum adenosine deaminase activity compared to controls (controls = 18.96 ± 4.61 U/l; typical = 25.09 ± 10.98 U/l; clozapine = 30.32 ± 10.83 U/l; $p < 0.05$, ANOVA) and 5′-nucleotidase activity was also increased in patients on clozapine. After adjusting for confounding factors, adenosine deaminase, but not 5′-nucleotidase, alterations remained significant particularly in the clozapine group. This result suggests that either altered adenosine metabolism is present in schizophrenic patients or is influenced by treatment with antipsychotics, particularly clozapine.

Keywords: Schizophrenia; Adenosine; Adenosine deaminase; 5′-Nucleotidase; Purinergic system; Antipsychotics

The neuromodulator adenosine has been recently proposed to contribute to the pathophysiology of schizophrenia [19,20]. This hypothesis postulates that a dysfunction in adenosinergic activity in schizophrenia would lead to putative alterations of dopaminergic and glutamatergic activities. Moreover, the ubiquity of adenosine could also account for some of the systemic alterations reported in schizophrenic patients [19,20,14,8]. The proposed adenosine dysfunction in schizophrenia, leading to a synaptic adenosinergic deficit, could be due to receptor alterations or altered metabolism, i.e. decreased production/release or increased degradation/uptake of adenosine [19]. This model is supported by indirect neurophysiological evidence such as sensory gating deficits in the P50 suppression paradigm induced by the adenosine receptor antagonists theophylline and caffeine in normal volunteers, resembling findings in schizophrenic patients [10,11]. Regarding pharmacological treatment, adjunctive treatment with dipyridamole, an adenosine transporter blocker, was beneficial compared to placebo for schizophrenia [2] and add-on treatment with allopurinol, which reduces purines degradation, was effective as adjuctive treatment in patients with poor response to antipsychotics [4,18] and in acute patients [1]. Finally, chronic treatment with clozapine, but not haloperidol, increased striatal ecto-5′-nucleotidase in rats [21].

Extracellular adenosine levels, and consequently the degree of receptor activation, depend on the rate of formation, diffusion and degradation of adenine nucleotides (ATP, ADP and AMP) and the nucleoside adenosine [3]. AMP formed from the degradation of released ATP can be hydrolyzed to adenosine by the action of 5′-nucleotidase, the rate-limiting step in the ectonucleotidase chain [26] (Fig. 1). Adenosine can then be either uptaken by nucleoside transporters or deaminated to inosine by adenosine deaminase [9]. Once inside the cell, adenosine can be converted to AMP by adenosine kinase or deaminated to inosine by adenosine deaminase (ADA) [3].

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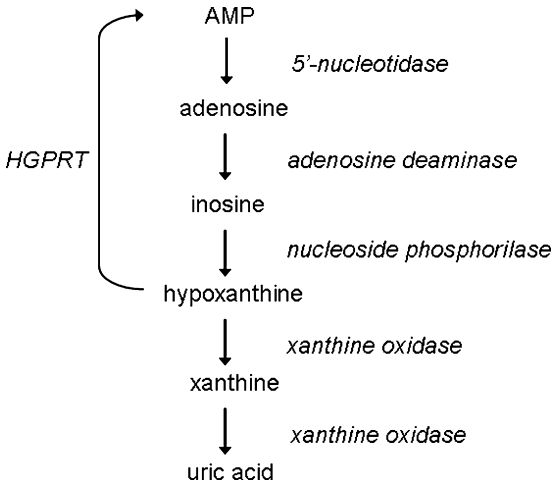

Fig. 1. Adenosine and its metabolism. Enzymes are in italics. HGPRT, hypoxanthine-guanine-phosphorybosyl-transferase.

In this study, we focused on peripheral adenosine metabolism by determining the activity of serum adenosine deaminase and 5ĭ-nucleotidase in schizophrenic patients under antipsychotic monotherapy with typical antipsychotics or clozapine and healthy control volunteers.

The protocol was in accordance to the Declaration of Helsinki and approved by Institutional Review Board of Hospital de Clínicas de Porto Alegre. All patients and a relative, as well as controls, provided written informed consent. The patients were selected from a specialized outpatient schizophrenia program from a University Hospital and their diagnosis had been confirmed longitudinally. Twenty-six male outpatients (18–52 years old) with a DSM-IV diagnosis of schizophrenia on antipsychotic monotherapy (14 on typical antipsychotics and 12 on clozapine) and 26 controls were studied. To be included, patients should have no other DSM-IV diagnosis in axis I based on clinical interview, no history of drug abuse (except nicotine) or use in the last month and no current medical condition. Controls balanced for age and race were recruited from the university and hospital staff and had no personal history of any axis I diagnosis, no family history of schizophrenia and no first degree relative with any psychotic disorder. The symptoms of all patients were evaluated with the PANSS [15].

Five (5) millilitre of peripheral blood was obtained by venous puncture with vacuum tubes without anticoagulants. Serum was separated by centrifugation and frozen to −70 °C until enzymatic assay. All blood samples were obtained between 4 and 6 p.m. Since 5′-nucleotidase activity is used as a liver dysfunction marker [22], liver enzymes AST, ALT, γ-glutamyltransferase and alkaline phosphatase as well as uric acid levels (the end-product of purines) were determined for control.

Serum adenosine deaminase activity was measured by a dry chemical technique (Vitro 75° Johnson & Johnson, USA), using reflectance for final determination [7]. Serum 5′-nucleotidase was measured using the 5′-nucleotidase Sigma kit (St. Louis, MO, USA) and processed in a RA XT-Bayer automatic equipment. All samples were analyzed in duplicate. Intra-assay variability was less than 10% for both assays.

Between-group comparisons were performed by *t*-test or factorial ANOVA for independent sample characteristics, as appropriate. Levene's test was used to evaluate data distribution. Enzyme activities and uric acid levels were compared between controls and patients on typical antipsychotics or clozapine with ANOVA, followed by Duncan test. Liver function tests were included as potential confounding factors using a linear regression model. The significance level was determined as <0.05. Analyses were performed with SPSS 10.0 software.

There were no differences between patients and controls regarding age and race-ethnicity, as well as between patients groups regarding age of onset or number of hospitalizations. Liver function tests were slightly increased only in patients taking clozapine, being statistically significant for alkaline phosphatase (Table 1). Schizophrenic patients on typical antipsychotics were on treatment with haloperidol, chlorpromazine or trifluoperazine.

Schizophrenic patients treated either with typical antipsychotics or clozapine presented increased serum adenosine deaminase activity compared to controls (controls = 18.96 ± 4.61 IU/l; typical = 25.09 ± 10.98 IU/l; clozapine = 30.32 ± 10.83 IU/l; $p < 0.05$, ANOVA, Duncan post hoc test), with no significant differences between the two groups of patients. After analysis using a multiple linear regression model with AST, ALT, alkaline phos-

Table 1
Clinical and laboratory sample characteristics

	Controls ($n = 26$)	Patients/typical antipsychotic ($n = 14$)	Patients/clozapine ($n = 12$)	Significance level (p)
Age	31.3 (±7.2)	34.9 (±9.0)	31.0 (±6.2)	0.31
Age of diagnosis		21.6 (±6.7)	18.1 (±4.6)	0.26
No. of hospitalizations		4.2 (±5.0)	7.3 (±7.1)	0.45
Total PANSS		62.5 (±21.5)	62.5 (±14.3)	1.00
Drug doses[a]		596 (±331)	518 (±183)	
AST	23.8 (±5.9)	21.1 (±6.1)	25.0 (±7.6)	0.29
ALT	32.4 (±9.6)	31.7 (±10.3)	41.1 (±17.5)	0.09
Alkaline phosphatase	69.0 (±11.5)	78.3 (±16.6)	88.0 (±30.6)[*]	0.02
γ-Glutamyltransferase	15.9 (±8.3)	15.6 (±5.5)	21.7 (±10.2)	0.09
Uric acid	5.6 (±0.8)	5.4 (±1.0)	5.5 (±1.3)	0.89

[a] Chlorpromazine equivalents.
[*] $p < 0.05$ compared to control group.